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Improving kettle holes as habitat and reproduction areas for amphibians – a case study in organic farms in north-eastern Germany

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Abstract. Kettle holes are found in young moraine landscapes and serve as an important habitat for amphibians. The loss of amphibians has been dramatic in recent decades, mainly because of the increase in land use intensity and deterioration of habitats e.g., kettle holes in agricultural landscapes. We monitored amphibian species on three organically managed farms in north-eastern Germany to get an overview of their occurrence and proof of reproduction to develop effective protection strategies. From 2016 to 2020, we investigated 50 kettle holes in cultivated fields. In 2018, we implemented the nature conservation measure 'cutting back dense wooded belts' in six of these kettle holes. Here, we focused on seven species considering four highly endangered species. We found six to seven species in up to 17 kettle holes in the 44 kettle holes without the measure 'cutting back dense wooded belts'. *Bombina bombina* occurred at the most kettle holes (57%). The number of kettle holes where amphibians reproduced differed strongly. On average, at least one species reproduced at 58% of the kettle holes. Many kettle holes become overgrown with negative effects for amphibians due to the reduction in solar irradiation and higher water consumption. The nature conservation measure increased the number of species on average from two to four and the number of species with reproduction from one to three. It is one of more than 100 measures in the 'Farming for Biodiversity' project that farmers can choose to receive a nature conservation certificate, which can be used for marketing purposes.

Keywords: agricultural landscape, *Bombina bombina*, clearing woody plants, *Hyla arborea*, monitoring, *Pelobates fuscus*, reproductive success, *Triturus cristatus*.

Introduction

Many amphibian species live in agriculturally managed regions and rely on water bodies for their reproduction (Kretschmer et al., 1995). They need diverse land habitats during the summer and winter as well as suitable migration corridors between these partial habitats (Wegener, 1991; Blab, 1993; Schneeweiss, 1996; Joly et al., 2001; Fischer et al., 2015; Glandt, 2018). The deterioration or destruction of these habitats and the intensive land use close to water bodies are the main causes for the decline in the amphibian populations (Günther, 1996; Jedicke, 1997; Mann et al., 2009; Lenhardt et al., 2015). New causes of danger are climate change and the increased incidence of fungal and viral diseases (Ohst et al., 2006; Mutschmann and Schneeweiss, 2008) as well as the release of non-native species (Kühnel et al., 2009) and the removal of animals for garden ponds (Schneeweiss, 2009).

In recent decades, there has been a sharp decline in many of the 21 amphibian species native to Germany (Günther, 1996). Twenty five percent are considered to be vulnerable, and a further ten percent are even endangered (BfN, 2015). Within the European network of protected areas NATURA 2000 (Raths et al., 2006), the amphibian species fire-bellied toad (*Bombina bombina*), yellow-bellied toad

(*Bombina variegata*), great crested newt and Italian crested newt (*Triturus cristatus*, *Triturus carnifex*) are listed as species requiring designation of 'Special Areas of Conservation' (Habitats Directive, Annex II). Other species, such as the common spadefoot toad (*Pelobates fuscus*), European tree frog (*Hyla arborea*), natterjack toad (*Epidalea calamita*) and European green toad (*Bufotes viridis*), are species in need of strict protection (Habitats Directive, Annex IV). In Brandenburg, nine species are on the Red List (categories 2 and 3) (Schneeweiss et al., 2004), in Mecklenburg-Western Pomerania 14 species are assigned to categories 1-3 (Kühnel et al., 2009).

Kettle holes are found in young moraine landscapes formed by the Weichselian or Wisconsin glaciation and are an outstanding scenic attraction of the agricultural landscape (Dreger and Luthardt 2006). They are described as 'eyes of the landscape' (Bergmann, 1983) and fulfil diverse functions within the agrarian area. Besides water storage and being a compensation zone for the hydrologic balance of the landscape, they also serve as a habitat for many plants and animals (Schneeweiss, 1996; Dreger and Luthardt, 2006; Lewis-Phillips et al., 2020; Pätzig and Düker, 2021). Land consolidation and changes in land management have often destroyed kettle holes in these areas (Kalettka, 1996). In the past, woody plants around small water bodies were used, for example, as firewood (Hamel, 1988). Nowadays, the branches of trees around kettles holes are only cut back when they grow into agricultural land. As a result, many kettle holes have become overgrown with reeds and woody plants, which, in addition to unwanted shading, can also lead to increased dessication due to increased water consumption (Pätzig and Düker, 2021). As overgrown kettle holes increase, they will lose their ecosystem function and services (Sayer et al., 2012; Pätzig and Düker, 2021). Species requiring higher water temperatures, such as *H. arborea* or *B. bombina*, can only reproduce poorly or not at all (Fog, 1996; van Buskirk, 2005; Helmecke, 2010; Pätzig and Düker, 2021). Therefore, the direct management of kettle holes by clearing the woody plants is recommended by various authors (Kalettka, 1996; Hamel, 1999; Skelly et al., 2005; Dodd, 2010; Kniep, 2010; Glandt, 2018; Clevenot et al., 2018; Pätzig and Düker, 2021). However, only three studies investigated the direct effects of woody plant removal/pruning and found positive effects for amphibians (Skelly et al., 2005; Werner et al., 2007; Helmecke, 2010).

In our study, we focused on two objectives: (i) to identify the most suitable kettles holes for nature conservation measures to support the occurrence and reproduction of amphibian species. (ii) to gather information about the effect of 'cutting back dense wooded belts' on the occurrence and reproduction. Beginning in 2016, we monitored a total of 50 kettle holes in the two study areas for five years. We expected a high variation of amphibian species and reproduction success which should offer valuable information for future recommendations for the implementation of targeted nature conservation measures on selected fields and kettle holes. On the basis of the first two investigation years, we selected 12 kettle holes from the 50 being monitored to test the effects of cutting back woody plants in the south-facing sections from 2018 to 2020. For both indicators, amphibian occurrence and reproduction, we expected positive effects with this nature conservation measure even for threatened species. The investigations are part of the accompanying scientific research in the 'Farming for Biodiversity' project (Gottwald and Stein-Bachinger, 2018). Organic farmers and WWF Germany initiated the project with the overall aim of raising the value of and increasing biodiversity on organic farms. At the same time, these benefits can be used for marketing purposes.

Materials and methods

Study area

Monitoring research on amphibian species took place on three organic farms in north-eastern Germany (federal states Mecklenburg-Western Pomerania (two farms, study area 1) and Brandenburg (one farm, study area 2)) (fig. 1). The study areas differ very slightly in terms of site and management conditions. The landscape is richly structured and diverse. Ground and terminal moraines with numerous kettle holes, marshy depressions and several larger lakes characterize it (supplementary fig. S1). Annual precipitation in study area 1 is 566 mm, and 569 mm in study area 2 (DWD, 2021). Table 1 shows the differences in precipitation distribution during the five study years. The climatic water balance in the months from April to September is generally negative. The soils consist of sand and sandy loams. The farms have been certified according to the EU and Biopark organic standards for more than 20 years. They raise suckler cows as important production segment (max. 0.5 LU/ha) besides producing arable cash crops. The farm in Brandenburg cultivates 3 347 ha (596 ha grassland, 2 751 ha arable land). The farms in Mecklenburg-Western Pomerania are situated directly next to each other. They cultivate 768 ha (475 ha grassland, 293 ha arable land) and 1 003 ha (353 ha grassland, 650 ha arable land).

Study design

In total, we monitored 50 kettle holes from 2016 to 2020 (28 on the two farms in Mecklenburg-Western Pomerania, 22 on the farm in Brandenburg) to get an overview on species richness of amphibians and proof of reproduction.

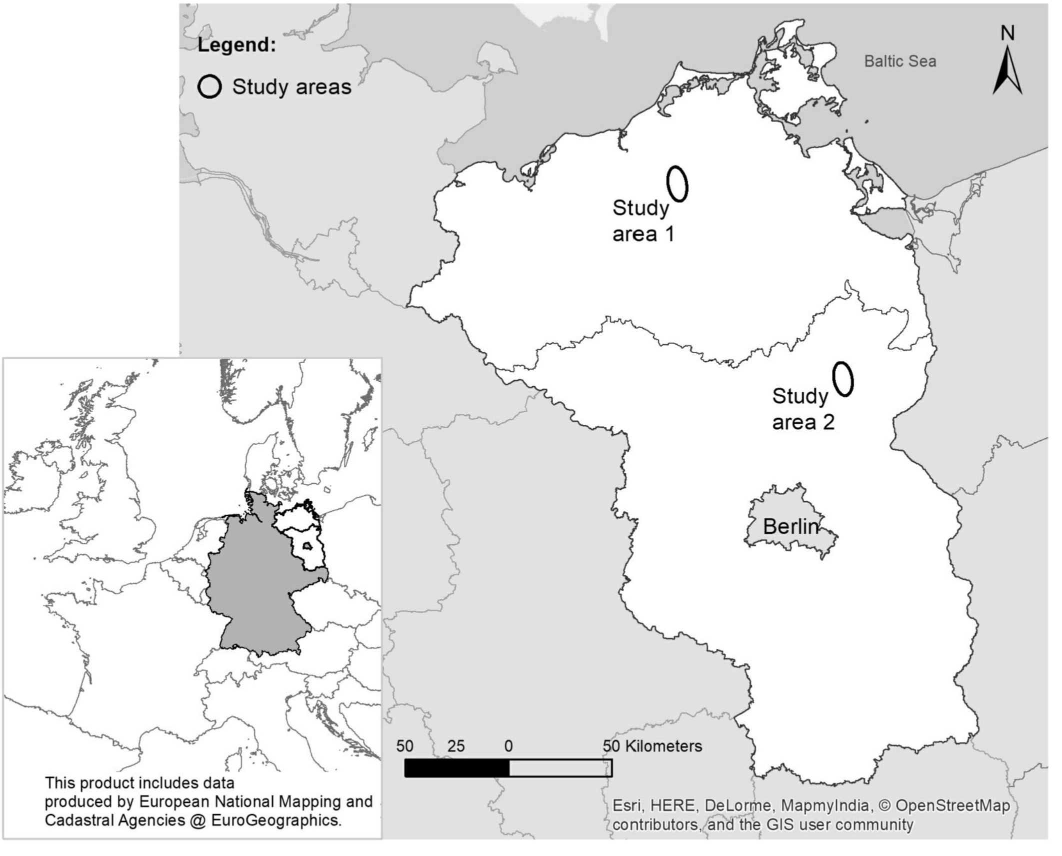

Figure 1. Location of the two study areas in Mecklenburg-Western Pomerania (study area 1) and Brandenburg (study area 2), Germany.

Table 1. Annual precipitation (mm) in the two study areas 2016 to 2020.

	2016	2017	2018	2019	2020	Mean 1991-2020
Study area 1	473	777	399	462	556	566
Study area 2	464	764	416	578	457	569

Table 2. Characterization of 50 investigated kettle hole types (Hamel, 1988).

Basic type	Characteristics	50 kettle holes
Open type (I)	Only open water body	7
Margin type (II)	Distinct water body	24
	Amphibian/terrestrial species with margin < 75 % of the water body area	
Reeds/reed bed type (III)	No distinct water body	7
	Amphibian/terrestrial species with reeds > 75 % of the water body area	
Woody type (IV)	Dominance of woody plants around and in the centre of the water body.	12
	No open water body recognizable or fully shaded	

The kettle holes can be assigned to four types (open, margin, reed, woody type) (table 2). In an experiment, we tested the effects of cutting back woody plants in the south-facing sections of 6 kettle holes. A further 6 kettle holes served as references (supplementary fig. S2). Before implementing the measure, in four cases the kettle holes (woody type) were not used for breeding anymore. In two cases (margin type), over 90% of the water surface was blocked from sunlight by the shadows cast by the trees. Amphibian species were still present with only partially declining reproduction or suitable habitat structure for the prioritized species. In February/March 2018, habitat improvement measures (clearing of woody plants with chainsaws up to 200 m^2 in the immediate riparian area and the water surface in the south-facing sections) were carried out on 2 kettle holes in Brandenburg and 4 in Mecklenburg-Western Pomerania. Four kettle holes of the same type were selected as references to the four kettle holes from woody type in which the clearing of woody plants took place, and the same was done for margin type. It was also important that similar numbers of amphibians were present in the kettle hole pairs prior to the measure. This was ensured by the surveys conducted in 2016 and 2017. The distances between the kettle hole pairs ranged from 130 to 900 m, with one exception. One comparison pair in study area 1 were 5 000 m apart. The size of the total of 12 kettle holes including the riparian area was on average 1 500 m^2 in study area 1 and 900 sqm in study area 2. To ensure comparability, care was also taken to ensure that both the surrounding landscape and the management of the agricultural land on which the kettle holes are located were similar. The water flow of the kettle holes was visually assessed on the basis of four types at each time of the survey (flooded: water flow over the edge of the water body, good water flow in comparison to significantly low water flow and dried-up: no longer relevant for the reproduction of amphibians).

Survey methods

We investigated all amphibian species known to be present in the regions (crested newt (*Triturus cristatus*), fire-bellied toad (*Bombina bombina*), common spadefoot toad (*Pelobates fuscus*), European tree frog (*Hyla arborea*), common newt (*Lissotrition vulgaris*), moor frog (*Rana arvalis*), European toad (*Bufo bufo*), common water frog (*Pelophylax kl. esculentus*), and European common frog (*Rana temporaria*)). A focus was on four particularly endangered target species: *T. cristatus* and *B. bombina* (Habitats Directive Annex II, IV), *P. fuscus* and *H. arborea* (Habitats Directive Annex IV).

In addition to watercourse surveys (e.g., to estimate water levels), spawn clump counts, acoustic surveys and transect surveys of riparian margins and wet areas near watercourses were carried out. The focus was on gathering information about the occurrence of species but not about the number of individuals. Net and light traps (Krone, 1992) were conducted to survey caudate amphibians and provide evidence of reproduction of all species. All kettle holes were surveyed five times per year from March to September during the five investigation years (2016-2020).

Statistical analysis

Data determines only the absence or presence of a species in kettle holes per year and whether reproduction took place. Chi-squared-tests were used to determine the differences between the four categories of kettle holes separately for every year (table 2).

For the experiment (2018-2020), a two-way ANOVA was used to assess the influence of cutting back dense wooded belts around kettle holes (= factor 1) considering annual effects as an important factor (2) on the number of species and their reproduction. Due to the limited numbers and heterogeneity, statistical tests were used to describe the effect of the nature conservation measure.

Results

Occurrence of species and reproduction success

A total of nine species were detected at the kettle holes. *Bufo bufo* and *R. temporaria* are two species that occurred in very low numbers (on average *Bufo bufo* in 5 kettle holes with reproduction in only 3 kettle holes during 5 investigation years, while *R. temporaria* occurred in one kettle hole in only three years with reproduction only in one year) because these habitats do not meet the typical habitat requirements of these

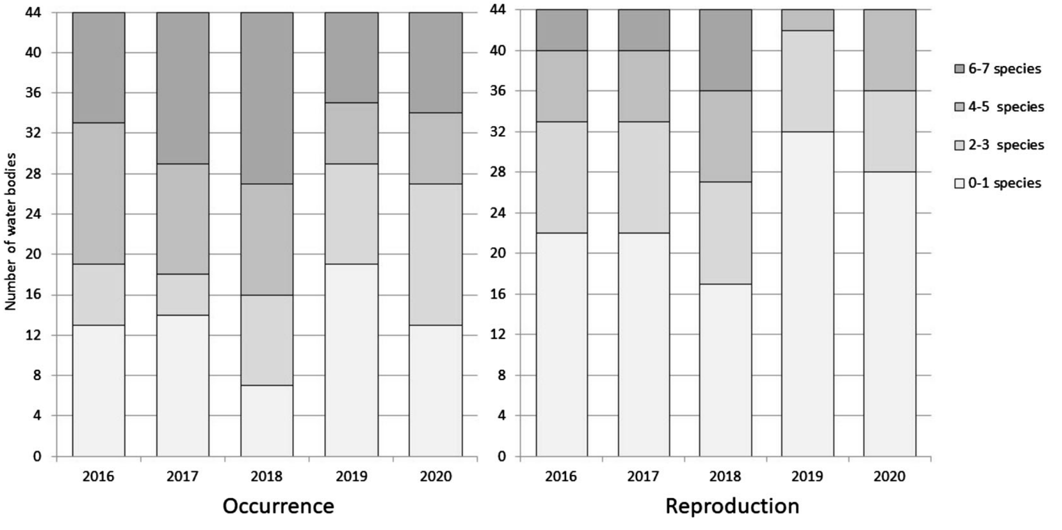

Figure 2. Number of kettle holes with occurrence (left) and reproduction (right) of amphibian species on organically farmed fields in north-eastern Germany ($n = 44$ kettle holes, 2016-2020).

species. For this reasons, they were not included in the analysis.

From 2016 to 2020, 6 to 7 species were present in 9 (2019) to 17 (2018) kettle holes (fig. 2). The highest numbers of occurrence were recorded in 2017 (15) and 2018 (17). The number of kettle holes where amphibian reproduction occurred varied greatly in individual years. The highest number of kettles holes (8) with reproduction of 6 to 7 species was recorded in 2018.

The occurrence and reproduction of each species over the five study years is shown in figure 3. On average, all 7 species occurred at 5 (11%) of the kettle holes during these years. The greatest variation in occurrence was seen for *P. fuscus* (at 9-32 kettle holes) and *R. arvalis* (at 9-25 kettle holes) (fig. 3 and supplementary table S1). While overall reproduction was low for *H. arborea*, the largest number of kettle holes with evidence of reproduction for *P. fuscus* and *L. vulgaris* were found in 2018 (supplementary table S1). Over the five years of the study, the highest numbers were recorded for *P. esculentus*.

At least one species was detected at 80% of the kettle holes, and at 67% of these, at least one priority species was detected over the mean of the five study years (supplementary table S1). Among the priority species, *B. bombina* occurred at the most kettle holes (57%). On average, at least one species reproduced at 58% of the kettle holes and at least one priority species reproduced at 40% of the kettle holes. With regard to the occurrence of the priority species, there was evidence of the highest reproduction for *T. cristatus* in 57% of cases, while for *H. arborea*, the reproduction was only 25% (supplementary table S1).

The various types of kettle holes differed significantly in the number of amphibian species that both occurred and reproduced there (fig. 4). Highest species numbers were occurring in reed type kettle holes (49% with 6 to 7 species), and least species occurred in woody type kettle holes. Despite a high variation, differences between types of kettle holes were found to be significant in four of the five study years for occurrence ($P < 0.05$; except 2018), and only in 2017 and 2020 for reproduction ($P < 0.01$).

Effects of the clearance of wooded belts on the occurrence and reproduction of amphibians

After cutting back the wooded belts at the 6 kettle holes, the type of succession at a total of

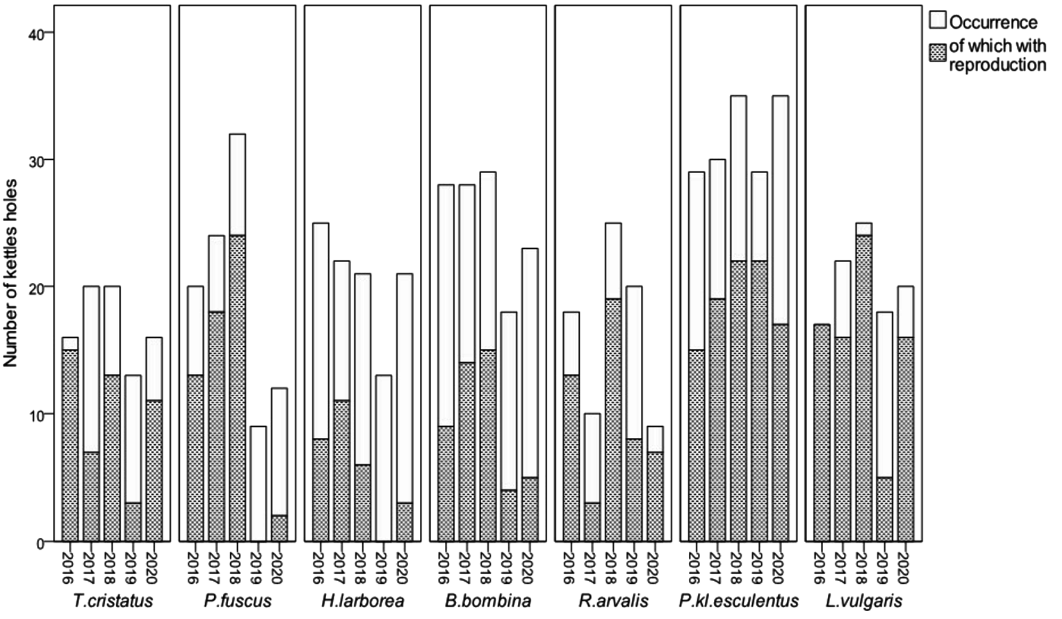

Figure 3. Number of kettle holes with single species occurrence and reproduction on organically farmed fields in north-eastern Germany ($n = 44$ kettle holes, 2016-2020).

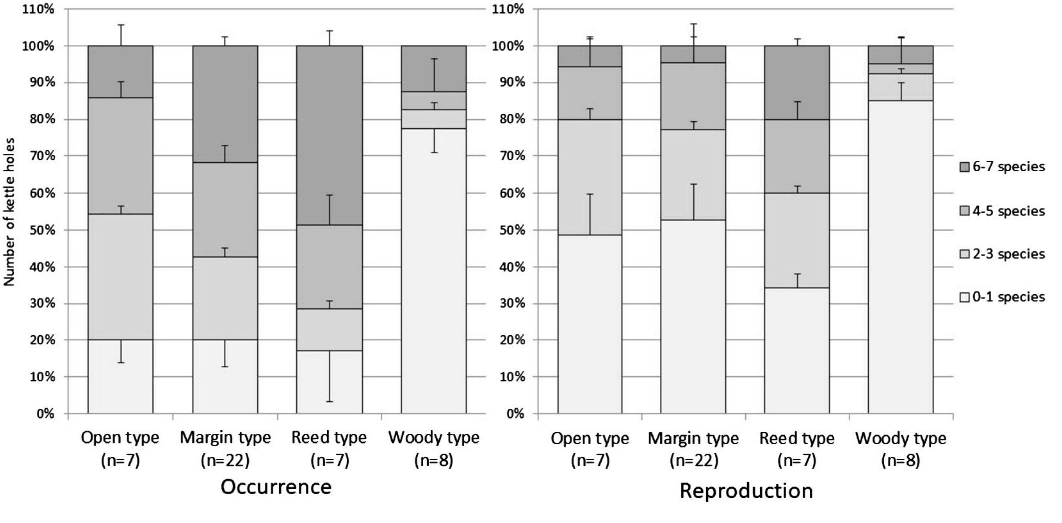

Figure 4. Preferences of the different amphibian species for four types of kettle holes (I to IV) with occurrence (left) and reproduction (right) (standard error, n = number of kettle holes, 2016-2020).

3 kettle holes changed from woody type to reed type. For one woody type kettle hole, an open water area was present after the measure, but due to its size, it remained in the woody type category. The same is true for both of the margin type kettle holes.

Considerably more species were detected after the clearance of the wooded belts ($P < 0.01$; Fig. 5a), as well as significantly positive effects on reproduction ($P < 0.05$; fig. 5b). If the four priority species are considered separately, there was a tendency for a positive effect

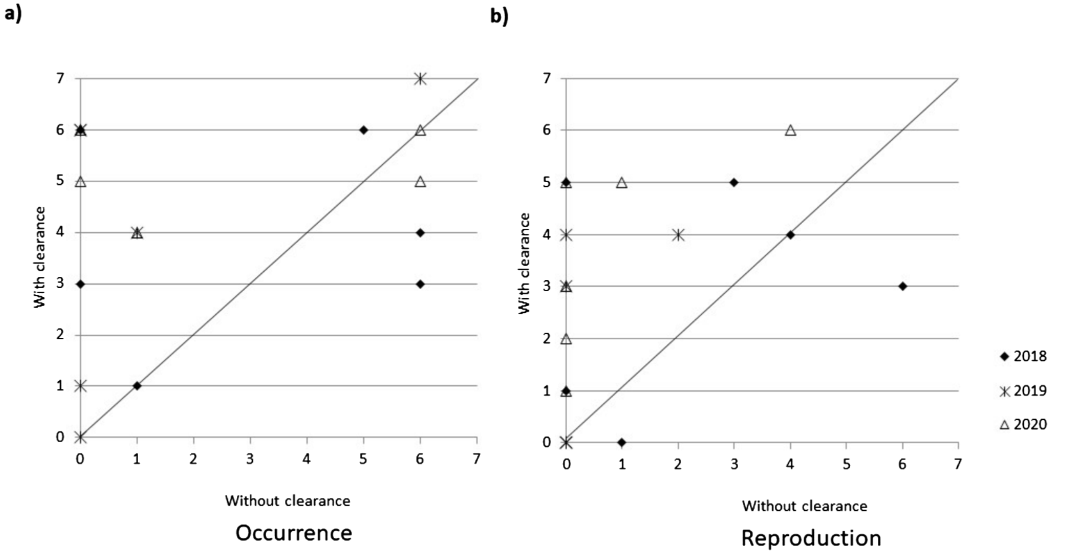

Figure 5. Influence of the clearance of woody plants on the number of kettle holes with species occurrence (a) and species with reproduction (b) in organically farmed fields in north-eastern Germany. 6 pairs of kettle holes are shown each with a maximum of 7 species (2018-2020). Each point represents a pair of kettle holes. The diagonal line describes no difference in species numbers, and points above show an incidence of more species after the removal.

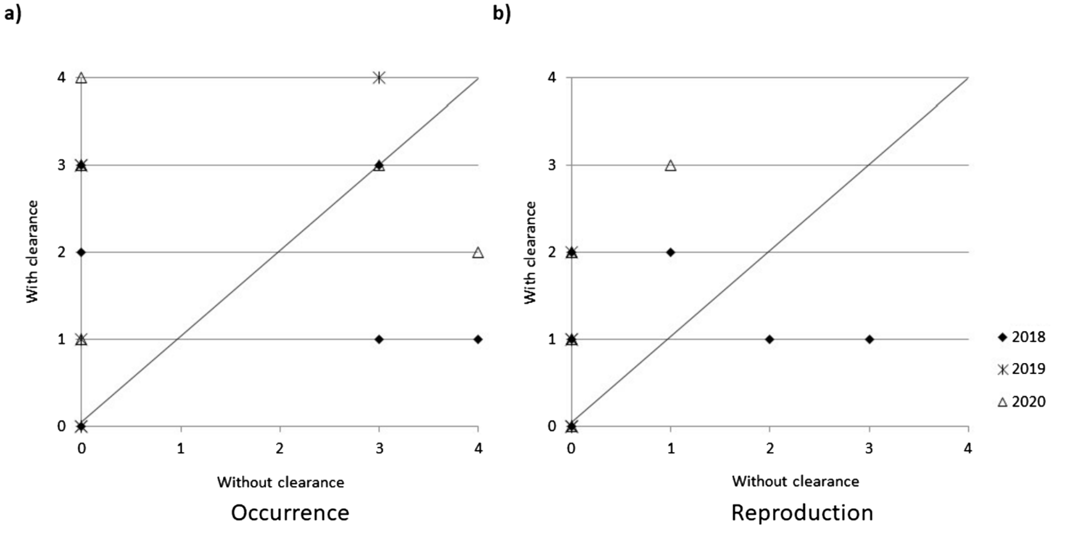

Figure 6. Influence of the clearance of woody plants on the number of kettle holes with priority species occurrence (a) and priority species with reproduction (b) in organically farmed fields in north-eastern Germany. 6 pairs of kettle holes are shown each with a maximum of 4 priority species (2018-2020). Each point represents a pair of kettle holes. The diagonal line describes no difference in species numbers, and points above show an incidence of more species after the removal.

on occurrence (Fig. 6a) and a significant positive effect on reproduction ($P < 0.05$, fig. 6b) after the clearance of woody plants. Strong annual effects are also clearly visible but not statistically significant.

Discussion

The agricultural study areas in Mecklenburg-Western Pomerania and Brandenburg are characterized by a large number of kettle holes.

They thus might offer good habitat conditions for amphibians, which depend on spawning waters for their reproduction. In study area 1, 9 of the total of 14 amphibian species occurring in Mecklenburg-Western Pomerania (Kühnel et al., 2009) were detected, while in study area 2 a total of 8 of the 15 species occurring in Brandenburg (Schneeweiss et al., 2004) were found in the investigated kettle holes. The relatively frequent occurrence of the four target species can be interpreted as a sign of good habitat quality and a structurally rich landscape.

Both study areas are located in major distribution areas of *B. bombina* in Germany (Schober, 1986; Schneeweiss, 1993). *T. cristatus* and *P. fuscus* are widespread in Mecklenburg-Western Pomerania, and in Brandenburg these species have their main population distribution within Germany (Schiemenz and Günther, 1994). *H. arborea*, *R. arvalis*, *L. vulgaris* and *P. esculentus* are found all over the country (Schiemenz and Günther, 1994). Nevertheless, all populations of the species have suffered from a massive decline in Germany and the four priority target species (*H. arborea*, *P. fuscus*, *B. bombina*, *T. cristatus*) are among the vulnerable or endangered Habitats Directive species. The investigated kettle holes are located in organically managed fields. Organic farming does not use mineral nitrogen fertilizers or synthetic pesticides which can either harm or kill amphibians when exposed to these chemical agents, as described by various authors in studies on conventional areas (Dinehart et al., 2009; Edge et al., 2011; Berger et al., 2011).

The reproductive suitability of small kettle holes for amphibians is quite crucially dependent on their water volume (Kalettka, 1996; Pätzig and Düker, 2021). The year 2017 was characterized by a particularly wet year for north-eastern Germany. The kettle holes were well filled with water. In 2018, despite lower precipitation, there was still sufficient water in the kettle holes. In the two subsequent years, some kettle holes dried up because of low precipitation. In 2020, 46% of the investigated kettle holes in study area 1 fell dry by September, including three of the woodland maintenance and reference kettle holes (37%). In study area 2, 86% of the kettle holes and all of the woodland maintenance and reference waters were dry in autumn. Increased bush encroachment of kettle holes will be accelerated under drier conditions (Sayer et al., 2012; Pätzig and Düker, 2021). A dense wooded belt around kettle holes decreases their quality as a habitat for amphibians due to the reduction in solar irradiation with negative effects on the development of the larva and their survival rates (Helmecke, 2010). In order to achieve the desired effect of increasing the exposure of the kettle holes to sunlight, the wooded areas must be removed in particular from the south side of the kettle holes. The positive effects on *H. arborea* and *B. bombina* identified by Helmecke (2010) were corroborated by the present results and this trend is also reflected for another five species studied. The positive effect of the clearance of woody plants in our studies for the four priority species was more evident in the reproduction than in the pure occurrence of the species. In addition to an improvement in the exposure to sunlight, removal of woody plants as conservation action leads to a lower consumption of water by these plants and subsequently to a higher water level of the kettle holes. Accordingly, in 2019, all reference kettle holes in both study areas showed a low water level as early as April or had dried out. In 2020, this was not so severe in study area 1, as opposed to study area 2, and there was no reproduction.

The positive effects of the nature conservation measure also depends on the quality of the clearance of woody plants. In kettle holes with a vegetation consisting mainly of willows and alders, the removal must be carried out directly at the bottom of the hole. In all cases, removal of the cuttings is required to prevent sedimentation in the water body. Tree prunings can be used as firewood. Such work can be easily planned and accomplished by a farm during the winter months. However, the clearing of woody plants at kettle holes can have negative effects on butterfly species or birds associated with woody vegetation. For example, the lesser purple emperor butterfly (*Apatura ilia*) needs sunny woody edges with poplars (*Populus tremula*, *P.* × *canadensis*) to lay its eggs and also likes to colonize field copses (Richert, 1999). In our study, however, the riparian areas were not cut back completely, so that this conflict can be assessed as very low. The impact on bird life is also low, since the woody plants were cleared during the vegetation dormancy and then only selectively (Hinsley et al., 2000). But these aspects should also be considered when restoring kettle holes.

The clearing of woody plants requires a permit since woody plants along water bodies are protected by law (§30 Federal Nature Conservation Act, BNatSchG, 2009). The application for a permit for water body maintenance should be made by landscape conservation associations or nature conservation consultants in order to relieve the burden on farmers. Overall, this nature conservation measure is an important contribution to the preservation and promotion of the herpetofauna with a relatively small amount of work.

There are additional factors that play a role on the distribution of amphibians in agricultural landscapes. Janin et al. (2012) detected an increase in stress hormone levels in the common toad while migrating over arable land. On organically farmed fields, as presented in this study, management operations like ploughing or mowing around water bodies can be a risk. Therefore, the establishment and extensive use of riparian strips, which can serve as summer habitat, is beneficial even on organic farms. Helmecke (2010) demonstrated in a long-term study on organically farmed areas in the vicinity of study area 2 that an increase in the number of riparian strips shifted the reproduction of *B. bombina* and *H. arborea* increasingly to the kettle holes with riparian strips. Therefore, riparian strips provide a further considerable improvement of the habitat for amphibians in summer and winter, if their management is adapted to the needs of the animals. This can be achieved if sensitive periods are considered, in which no management operations take place (e.g., first and second cut of leys should be before July and subsequent cut in October (Gottwald et al., 2010)).

In conclusion, the results of our long-term monitoring of the occurrence and reproduction of amphibian species in kettle holes provide valuable information where nature conservation measures or combinations of measures for improvement should be implemented. The study also highlights the importance of clearing woody plants on the south-facing sections of kettle holes on species diversity and reproduction success. The measure should be carried out without bureaucratic effort and be supported by environmental schemes to increase acceptance by the farmers. Wood pruning could also be used as wood chips and thus contribute to sustainable land use. The description of this measure as well as further nature conservation measures for amphibians is part of the service catalogue with more than 100 modules from which farmers can select measures to receive the ‘Farming for Biodiversity’ nature conservation certificate (Gottwald and Stein-Bachinger, 2016, 2018). Several measures are very effective on a small-scale, such as the removal of woody plants on the south-facing sections of kettle holes, the creation of riparian strips or green strips as a connection between two kettle holes. At the same time, other ecosystem services provided by kettle holes will be increased (Lewis-Phillips et al., 2020). In future studies, the focus should be on the effects of combinations of measures in order to best exploit synergies for amphibian protection. Since most of these measures are associated with additional expenses and yield losses for a farm and need to be compensated

(Ruehs and Stein-Bachinger, 2019), they should be implemented at selected kettle holes where they provide the greatest effect from a conservation point of view. Therefore, long-term monitoring of kettle holes in agricultural managed areas is required to investigate their ecological state and i.e., the succession process towards woody types to give specific recommendations for improvement. Especially the drought in the last three studied years can increase potentially irreversible effects with a future loss of kettle hole types.

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Supplementary material. Supplementary material is available online at:
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